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BACTERIAL MUTATION STUDY

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EXECUTIVE SUMMARY

The experiment presented in this document was a conceptual replication of reported work in the parapsychological literature, claiming positive statistical evidence for psychoenergetic interactions with biological systems. Both the energetic and informational aspects of human interaction with bacteriological systems were examined, with the ultimate objective of determining, to first order, whether biological systems can be employed as psychoenergetic "intrusion detectors."

There were two principal experimental hypotheses under consideration. The first, which will be referred to as the Intuitive Data Sorting (IDS) hypothesis, posits that individuals are able to identify or "sort out" locally-deviant subsequences contained within a larger random sequence using psychoenergetic means. In our experiment, an IDS hypothesis predicted that individuals would be able to identify psychically--from a set of test tubes with a normal statistical spread of mutation rate--subsets of test tubes either with slightly higher or slightly lower average mutation rates than the overall mutation rate for the entire set. Because an IDS mechanism appears to be predicated on an individual's ability to gain information about a system psychoenergetically, it is thought to involve *informational processes* primarily.

The second experimental hypothesis, which will be referred to as the Remote Action (RA) or IDS Unfavorable (IDSU) hypothesis, postulates that certain individuals are able to effect either a predetermined increase or decrease in a given sample's mutation rate, by somehow "mentally" causing physical (e.g., genetic) changes in the bacteria. Because an RA mechanism appears to be predicated on an individual's ability to effect physical changes in a system psychoenergetically, it is thought to involve *causal or energetic processes* primarily.

A total of seven subjects contributed six sessions each: three sessions were designed to test the IDS hypothesis, and three were designed to test the RA hypothesis. In all sessions, the subject was confronted with nine test tubes, which were visible inside a locked, environmentally-stable ice chest. The tubes contained dilute solutions of the bacterium *Salmonella typhimurium*. The bacteriological preparations were carried out by SRI's Microbial

Genetics Department, which routinely uses the Ames *Salmonella* assay that was adapted for use in this study.

In the IDS sessions, the subjects were able to choose three test tubes in which they wished to promote the mutation rates psychoenergetically (high aim), three tubes in which they wished to inhibit mutation rates (low aim), and three that they wished to leave "uninfluenced" as controls (no aim). In all of the RA sessions, Tubes 1, 2, and 3 were predetermined as the low-aim tubes (the subject would attempt to inhibit mutation rates); Tubes 4, 5, and 6 were the no aim controls; and Tubes 7, 8, and 9 were the high-aim tubes (the subject would attempt to promote mutation rates). The basic premise in comparing the IDS and RA conditions is that the subjects were given the opportunity to select high-versus-low mutation rates from a natural spread of nine in the IDS sessions. Given the predetermined tubes of the RA sessions, however, the subjects were required to cause physical changes in the bacteria, in order to achieve the desired high-versus-low mutation rates.

The overall result of the experiment showed weak statistical evidence that individuals are able to sort bacteriological samples according to mutation rate--that is, a $p \leq 0.05$ was obtained overall in the IDS sessions for the mutation rates of the low-aim test tubes being lower than the no-aim controls. Statistical significance was not achieved in any of the other IDS conditions (i.e., for no-aim mutation rates being less than high aim or for low aim being less than high aim). There were no significant differences for various aims observed in the RA condition. It must be concluded, therefore, that while there was some evidence that subjects are able to gain information psychoenergetically about the mutation rates of *Salmonella*, there was no compelling evidence that subjects are able to cause physical perturbations in these bacteria.

According to criteria set forth in the beginning of this study, a physical system will not be considered a candidate intrusion detector unless there is clear evidence that it is registering energetic effects (i.e., physical perturbations) concomitantly with psychoenergetic intent. To first order, therefore, it must be concluded on the basis of this one experiment that the *Salmonella* bacterium does not appear to be a promising intrusion detector.

Because this is the only known experiment of its kind using *Salmonella* bacteria as the target biological system, replication is strongly recommended--both to verify the

robustness of the IDS capability, and to evaluate definitively the efficacy of using *Salmonella* as an intrusion detector.

I OBJECTIVE

The objective of this subtask was to determine the veracity of the claims in the parapsychological literature regarding psychoenergetic interactions with biological systems. A conceptual replication of the most promising of these earlier claims was undertaken, as a means to examine whether biological systems register physical effects concomitantly with psychoenergetic "intent" by an observer. This initial experimental effort was an attempt to determine, to first order, whether biological systems can eventually be employed as psychoenergetic "intrusion detectors."

II INTRODUCTION

One of the ultimate applications goals of psychoenergetic phenomena is the determination of whether psychoenergetic intrusion can be detected, and whether countermeasures exist against such intrusion. From a phenomenological perspective, the term *psychoenergetic intrusion* can entail what appears to be either energetic or informational processes, or both, as indicated by the following set of operative definitions:

- The direct perturbation of physical systems that appear to be well shielded against, or otherwise inaccessible to, human influence (energetic).
- The psychoenergetic acquisition of information thought to be secure against access (informational).
- The perturbation of a physical system that occurs indirectly as a result of an individual's attempts to acquire information through psychoenergetic means (energetic and informational).

Only those intrusions that entail causal interactions with physical systems are likely to be detected. A physical system will not be considered a candidate intrusion detector, therefore, unless it registers energetic effects directly (as a result of intentional perturbation), or indirectly (as a result of concomitant acquisition of information).

In the parapsychological literature, the energetic manifestations of psychoenergetic intrusion are variously referred to as remote action (RA), remote perturbation (RP), psychokinesis (PK), telekinesis (TK), and so forth; informational processes are most often referred to as remote viewing (RV), clairvoyance, precognition, and the like. The term *countermeasures* may be defined as the shielding or jamming of psychoenergetic intrusion by either physical or mental processes.

Before the higher-order problem of countermeasures can be addressed, experimental verification of the existence of psychoenergetic intrusion must first be obtained. Detection of the putative energetic aspects of psychoenergetic intrusion can be accomplished most directly by designing experiments in which an individual's primary task is to actively attempt to cause perturbations in various types of physical systems. Numerous RA

experiments of this type, using a wide variety of physical systems, have been cited in the parapsychological literature.

One category of candidate target physical systems is biological systems; the precedent for using these in RA experiments has been well established. Of particular interest (because of its similarity to the experiment detailed in this document) is Carroll B. Nash's experiment involving the psychokinetic control of bacterial mutation.* The published abstract of the Nash experiment is provided here:

Three experimenters each tested 20 subjects not known to be psychically gifted. Because of procedural errors, results were obtained for only 52 subjects. Each subject was tested in a single run with a separate set of nine tubes of a mixed culture of lac-negative and lac-positive strains of Escherichia coli. Mutation of lac-negative to lac-positive was mentally promoted in three of the tubes, mentally inhibited in three, and three of the tubes served as controls. The mutant ratio of lac-positive to total bacteria was greater in the promoted than in the inhibited tubes, with two-tailed $p < 0.005$; less in the inhibited tubes than in the controls, with two-tailed $p < 0.02$; and greater in the promoted tubes than in the controls, although not significantly so. The results are interpreted to suggest that the rate of bacterial mutation was psychokinetically affected.

The experiment described in this report also undertook to investigate psychokinetic influence on bacterial mutagenicity, but it differs significantly from the Nash experiment in certain of its experimental protocols and underlying theoretical assumptions. The overall objective was also different than that of the Nash experiment in that the SRI study is concerned with providing a "first order" examination of the existence of psycho-energetic intrusion detection with biological systems, as a precursor to investigating the necessity and/or feasibility of countermeasures against such intrusion.

* Nash, C. B., "Psychokinetic Control of Bacterial Growth," *Journal of the Society for Psychical Research*, Vol. 51, pp. 217-226 (1982).

III METHODS OF APPROACH

In this chapter, we present a few concepts that are preparatory to a discussion of the actual protocols used in our experiment. The preliminary or background concepts are presented in two separate sections: "Definition of Terms," and "Biological Background." The third section, "Experimental Design," provides an overview of the experiment in terms of how it was designed to address the proposed theoretical concerns.* The fourth section, "Protocols," is a detailed summary of all presession, session, and postsession procedures.

A. Definition of Terms

In order to provide a framework for discussing the essential components of this experiment, working definitions of the most salient and most frequently encountered biological- and protocol-related terminology are provided below.

1. Biological Terms

The following are the most common biological terms:

- **Bacteria**--Microscopic, unicellular organisms used as the target biological system in this experiment. The specific species employed was *Salmonella typhimurium*, a short, rod-shaped bacterium that is actively motile.
- **Histidine**--An amino acid essential for the growth of *Salmonella typhimurium*.
- **Mutation**--A genetic change occurring in a small subset of *Salmonella* that allows them to grow and divide in the absence of histidine. Such cells are termed mutant cells or *revertants*.
- **Mutation Frequency**--Number of mutant cells per total number of cells--normalized, for comparison purposes, to number of mutants per million cells.
- **Plating**--A process whereby diluted samples of the bacterial cultures in the experimental test tubes were placed on complete medium plates (i.e., plates

* Our theoretical considerations have been fully addressed in the "Executive Summary."

with medium containing histidine), and selective minimal glucose plates (i.e., plates with medium lacking histidine) to allow for the growth and appearance of bacterial colonies.

2. (U) Procedural Terms

The following are the most common procedural terms:

- **Subject**--One of seven volunteers who undertook to psychoenergetically influence the mutagenicity of the bacterial samples.
- **Monitor**--The individual recording the events that transpired during an experimental session, and supervised the subject's activities.
- **Technician**--The microbiologist who was responsible for all aspects of the pre- and post-session preparation of the biological samples.
- **Session**--A single sitting in which the subject attempted to (1) increase the mutation rate of bacteria placed in three test tubes, (2) decrease the mutation rate of those placed in three different test tubes, and (3) leave yet a different group of three uninfluenced as "controls." Each of the seven subjects contributed six such sessions.
- **Trial**--An attempt by a subject to psychoenergetically influence (or not influence, as in the case of control test tubes) the bacterial culture in a single test tube. There were nine such trials in each experimental session.
- **Controls**--Two types of controls were employed in this experiment: intrasession and extrasession. Intrasession control test tubes consisted of three bacterial test tubes, which the subject was instructed **not** to attempt to actively influence, from among the set of nine session test tubes. Extrasession controls consisted of two tubes per session that were prepared by the technician in exactly the same manner as the session test tubes, but were not used as part of the experimental session set of nine tubes. The extrasession controls remained at all times in the Microbial Genetics Laboratory, and provided the requisite data for establishing an independent measure of mutation rate.
- **Feedback**--A drawing presented to the subject that indicated his/her performance on a given session. Feedback for a given session was typically administered prior to the start of the subject's next session.

B. Biological Background

In this section, we will give a general overview of the Ames *Salmonella* assay that is used routinely by SRI's Microbial Genetics Department, and that was adapted for use in this experiment to study psychoenergetic effects on mutation frequency.

The heritable material of living organisms is contained in the DNA (RNA in some viruses), a large molecule so constructed that it can replicate itself in a most exact fashion one cell generation after another. This is the basis of biological continuity and unity. It is also, however, the basis for biological diversity, which occurs through mutations. Each mutation alters the action of a specific gene, which is a genetic entity with its own specific end product, or protein. Genes are very stable structures, but each has its own spontaneous mutation frequency. The probability that a spontaneous mutant cell will be obtained every time a cell divides is constant, provided the environmental conditions are unchanged. Changes in the environment are known to influence the mutation frequency. Such changes include the presence or absence of certain trace elements (e.g., selenium), plus the presence of physical or chemical agents (mutagens).

Bacteria provide a convenient way to study mutations because millions of cells can be grown in a very short period of time. Over the past few years, several bacterial assays have been developed to screen chemicals for their ability to induce mutation. Because there is a close correlation between mutagenesis and carcinogenesis, such mutagenicity assays are very often used together with the *in vitro* tests that employ single microbial and/or mammalian cells, as well as *in vivo* tests that employ multicell organisms from insects (fruit fly) to mammals (rodents). One of the best known bacterial mutagenesis assays is the *Salmonella*/mammalian microsome histidine reverse mutation assay developed by Dr. Bruce Ames at the University of California in Berkeley. The Microbial Genetics Department at SRI International is using this assay system on a daily basis for Government agencies and commercial clients to determine the mutagenic potential of chemicals; they have performed such testing over a period of more than 10 years.

The *Salmonella* assay employs several tester strains of *Salmonella typhimurium*, each with a unique specificity for detecting chemical mutagens. The *Salmonella* strains, under optimum conditions, have a generation time of less than 30 minutes. The bacterial strains are unable to grow in the absence of the essential amino acid histidine because of a mutation in one of the genes that is needed for histidine synthesis. When these bacteria are plated on defined selective medium having little or no histidine, little or no growth occurs except those few bacteria that spontaneously mutate back to histidine independence (ability to grow in the absence of histidine). In this case, a nonfunctional gene product is reverted back to a functional one. This event allows the mutant cells to grow and divide. Because a mutation is stably inherited, all progeny of the mutated cells retain the ability to grow in the absence of

histidine. Distinct individual colonies will appear on the solid selective growth medium, with each colony containing billions of progeny of the spontaneously mutated cells. Exposure of the bacteria to a chemical mutagen will result in an increased number of colonies appearing on the solid selective growth medium, due to an increase in mutation induction.

In the Ames *Salmonella* assay, a small amount of histidine is added to the growth medium to allow for a few cell divisions of all the plated histidine requiring mutants ($\sim 10^4$). Such growth is often necessary for chemical mutagenesis to occur. The results of the *Salmonella* assay are usually expressed in terms of the number of revertant colonies per amount of chemical added to the selective growth medium, which is usually delivered to the plate in 25-ml volumes. Because of the presence of limited histidine in the selective medium, the results of the Ames *Salmonella* are considered "semiquantitative," since residual growth on all plates (control as well as chemical treated) does not allow for quantitative survival determination. A quantitative mutation frequency, however, can be determined. It is more labor intensive than the standard Ames assay, because survival determination requires diluting of the cell cultures, and a different growth medium is needed for determining (1) the mutant fraction and survivors for each of the controls, and (2) the different exposure concentrations of the test chemical. The mutation frequency is defined in terms of number of mutants per given number of surviving cells, usually per 10^6 cells.

Because of its simplicity and the rapid response time of about two days, the Ames *Salmonella* assay can readily be adapted to study the effect of RA on the mutation frequency. Such an adaptation was established by SRI's Microbial Genetics Department for use in this experiment; a detailed discussion of the specific biological procedures that were followed can be found in Section D, *Protocols*.

C. Experimental Design

1. Conceptual Replication

The experiment undertaken in this study represents a conceptual replication of the Nash experiment described in our Introduction chapter. The replication presented here is termed **conceptual**, because several of the experimental details of the Nash experiment have been changed and improved. First, two potential mechanisms have been postulated that could account for the acquisition of a statistically significant effect--that is, an IDS hypothesis has been advanced, in addition to the more established RA hypothesis. Second, *Salmonella*

typhimurium rather than *Escherichia coli* were used as the target bacterial cultures. Because this particular species of *Salmonella* is used most frequently by SRI's Microbial Genetics Department in toxicity studies, its behavior is particularly well understood in terms of assay conditions and experimental protocols. Finally, the Nash analysis was extended to include multiple analyses of variance.

As in the Nash experiment, nine test tubes filled with dilute bacterial culture were used per session. Mutation from histidine dependence to histidine independence was mentally promoted by the subject in three of the tubes, mentally inhibited in three, and the remaining three tubes served as controls. For the purposes of obtaining baseline data, two additional control test tubes (for a total of eleven altogether per session) were prepared in the same manner as the session test tubes, but were kept in the Microbial Genetics Laboratory.

2. Model Testing Criteria

a. The IDS Model

As mentioned previously, there were two primary models under investigation in this experiment. A pivotal concept to the first, or IDS favorable model, is **freedom of choice**: namely, that by using some type of psi-mediated informational processes, subjects have the opportunity to select out locally-deviant subsequences from a larger random sequence. For example, in half of the sessions, the subjects were allowed to select the three test tubes in which they wished to promote mutation, and the three test tubes in which they wished to inhibit mutation. A statistically significant deviation from mean chance expectation (MCE) in this condition, therefore, could be interpreted theoretically in two ways: (1) the subjects somehow mentally "forced" genetic changes to occur in the bacteria in accordance with their desires to either promote or inhibit mutation rates (the RA hypothesis); or (2) given the natural spread of mutation rates in a biological system, the subject was able to psycho-energetically sort those test tubes containing bacteria with high mutation rates from those tubes containing bacteria with low mutation rates (a session-by-session IDS hypothesis).

b. The RA or IDSU Model

The second model under investigation has been termed the Remote Action (RA) or Intuitive Data Sorting Unfavorable (IDSU) model. In this condition, the conduits by which either the subject or experimenter are able to select test tubes are rendered

as inaccessible as possible by the experimental protocols, such that the most direct explanation of a potentially significant effect is a remote-action mechanism. For the experiment presented here, the test tubes were fixed and predetermined as to aim for half of the sessions--that is, Test Tubes 1, 2, and 3 were *a priori* assigned as those tubes in which mutation rates were to be inhibited (low aim), Tubes 4, 5, and 6 were designated as controls (no aim), and Tubes 7, 8, and 9 were assigned as those in which mutation rates were to be promoted (high aim). In this way, the subject's potential IDS ability to sort tubes psycho-energetically according to the natural spread of mutation rates was precluded in half of the sessions.

A second possibility also had to be accounted for--namely, that experimenters, who are desirous of a certain outcome to the experiment, might use their own IDS ability to psychically scan the future. They could thus assign the IDSU sessions to days on which, for whatever reason (e.g. systematic sequential pipetteing bias in preparation of the cultures), Tubes 1, 2, and 3 might possess, on average, naturally lower mutation rates than Tubes 7, 8, and 9. To preclude this possibility, a balanced binary random protocol, with certain states disallowed, was formulated to determine session sequence. The disallowed sequences included (1) three RA sessions followed by three IDS sessions ("aabbbb"); (2) three IDS sessions followed by three RA sessions ("bbbaaa"); and (3) alternating IDS and RA sessions ("ababab" or "bababa"). The six-session sequence that was actually generated was RA, RA, IDS, RA, IDS, IDS (i.e., "aababb"), a protocol that was fixed for all subjects prior to the start of the experiment.

The final possibility that had to be considered was the degree to which the biological technician should be allowed to know the overall purpose of the experiment, in general, and to know specific experimental protocols, in particular. It was hypothesized that the technician's knowledge concerning experimental goals could potentially introduce his (the technician's own) IDS ability into the experiment as an undesirable variable. For example, had the technician been cognizant of the aims of the IDSU condition, it is possible that he could have used his own IDS capabilities either consciously or subconsciously to position the test tubes in the rack, such that Tubes 7, 8, and 9 possessed the high mutation rate cultures, and Tubes 1, 2, and 3 the low mutation rate cultures. Rather than precluding or, at the very least, greatly inhibiting IDS channels (in accordance with the original aim of its design) the IDSU condition would have created a new, unintended conduit by which IDS could operate directly. To preclude the potential introduction of this third possible IDS variable into the

experiment, the technician was kept blind as to all aspects of experimental goals and protocols until the conclusion of the experiment.

Given the caveats introduced by the experimental design considerations detailed above, a statistically significant deviation from MCE in the RA or IDSU condition could be interpreted, theoretically, as follows: (1) the subject somehow mentally "forced" genetic changes to occur in the bacteria in accordance with his desires to promote mutation in Test Tubes 7, 8, and 9, and inhibit mutation in Tubes 1, 2, and 3 (the RA hypothesis); or (2) IDS is able to operate in a goal-oriented mode, such that either the experimenter or the technician or both were able to adjust their actions in "real time" based on their global scan of a successful outcome of the experiment in the future (the global IDS hypothesis). To cite a specific example, if, at the conclusion of the experiment, the technician were finally told the purpose of the experiment, and that the result had been significant in the IDSU condition, there exists the following rather circuitous pathway by which his global IDS ability might be operative: (a) the technician could continually scan his future and determine that by pipetteing the cultures a certain way in real time, the IDSU condition would not show overall significance (that is, future "a" determines action "a" in real time); (b) he might discover, however, that by adjusting his real-time pipetteing strategy on certain sessions (e.g., by placing more dilute culture in Tubes 1, 2, and 3 as opposed to Tubes 7, 8, and 9), the IDSU condition would become significant overall (future "b" determines action "b" in real-time). It should be noted that the concept of global IDS is predicated on an "alternative futures" model, and it cannot be determined at present whether such a model possesses any validity. In any event, given the torturous nature of the pathway that must be postulated for global IDS to occur, we believe that a statistically significant result in the IDSU condition would be explained most parsimoniously by the RA hypothesis.

D. Protocols

1. Preexperiment Protocols

a. Subject Selection

A total of seven subjects were chosen primarily on the basis of their expressed interest in the field of psychoenergetics. None of the seven participants had ever taken part in a psychoenergetic experiment involving biological systems, and three of the group were true neophytes--having never participated in any type of psi experiment. Four of

the seven had been participants in previous psychoenergetic experiments--i.e., all four had demonstrated some ability in remote viewing. One of these four subjects also scored significantly in an earlier SRI random number generator PK experiment, and another had demonstrated some ability previously in Computer-Assisted Search (CAS) tasks. All of the participants were SRI employees: one was a statistician, two were secretaries, and the remainder were research professionals in either physics or computer science.

b. **Experiment Site Locations**

For reasons stated in Section C.2.a, it was determined that the biological technician should be kept entirely blind as to all facets of the experiment, and that, in order to facilitate this situation, the psychoenergetic testing should occur in a location that was different from the one used for the biological preparations. The Microbial Genetics Laboratory, therefore, was used for all aspects of preparation of the biological cultures, and a room in another building at SRI was used for the psychoenergetic sessions.

c. **Hardware Construction**

Once it had been determined that two separate facilities were necessary for conducting the experiment, a container had to be constructed that was suitable for transporting the biological samples from the Microbial Genetics Laboratory to the psychoenergetics facility. There were three primary factors that dictated the design of the container: (1) the biological samples had to be protected from extreme variations in temperature; (2) the samples had to be protected from sunlight; and (3) the container had to be lockable.

To control against extreme variations in temperature, which can greatly affect the mutagenicity of *Salmonella*, a Coleman® ice chest was chosen as the transport container. Triple-paned insulated glass windows were specially installed in the top and front side of the ice chest to allow an unobstructed view of the experiment test tubes. Because sunlight also affects the mutagenicity of the bacteria, a tarpaulin was used to completely cover the cooler during transport between the biological laboratory and the psychoenergetics facility. A lock was installed; the key was retained exclusively by the biological technician, to preclude the possibility of tampering with the biological samples once they had been removed from the Microbial Genetics Laboratory.

2. Presession Protocols

A series of activities took place prior to the start of every experimental session. First, the experiment monitor identified the session type from the fixed session sequence, "aababb," (cf. Section C.2.b), to determine whether the session would be an IDS favorable or an IDS unfavorable (RA or IDSU) condition.

Second, the technician in the Microbial Genetics Laboratory prepared the bacterial cultures for the session (see Appendix). Eleven numerically-labelled, sterile, 16- \times -150-mm test tubes were aseptically filled with 2.5 ml of glucose minimal broth. Fifty μ l of a 37°C overnight culture of strain TA100 of *Salmonella typhimurium* was then added to each tube.* In a standardized manner, the first nine tubes were arranged in a test-tube rack, which was placed in the specially designed ice chest, and then locked. The remaining two control cultures were shielded from visible light by a covering of aluminum foil, and were maintained at room temperature in the Microbial Genetics Department laboratory.

The ice chest, with its enclosed cultures, was placed on a cart and covered with a tarpaulin to ensure that the mutation rate of the cultures was not affected by sunlight during transportation from the laboratory to the experimental facility.

Third, the experiment monitor transported the covered ice chest on the cart from the biological laboratory to another facility at SRI, where the psychoenergetic portion of the experiment was performed. Prior to the arrival of the subject, the monitor wheeled the ice chest into a room equipped with a table and two chairs. The ice chest was then uncovered and positioned in such a way that a seated subject could readily view the nine numbered test tubes through the glass.

3. Session Protocols

For other than the first session for each subject, a session usually commenced with feedback to the subject of the previous session's results (to be discussed in "Postsession

*(U) It should be noted that there was no visible evidence of "cloudiness" caused by the bacterial culture in any of the prepared test tube solutions. The appearance of the liquid was uniformly that of clear tap water. Thus, there were no visual cues available to the subject, as to which test tubes might contain greater amounts of the bacterial culture.

Protocols" below). After the presentation of feedback, the subjects were informed whether they would select the test tubes they wished to influence (IDS favorable), or whether the tubes had been predetermined as to aim by the protocol (IDS unfavorable) for the current day's session.

For the IDS favorable session, the subjects were specifically instructed to select three test tubes that they thought were most susceptible to a decrease in mutation rate, three most susceptible to an increase in mutation rate, and three that they wished to leave uninfluenced as controls. It should be emphasized that in all cases where we use the term "select," we mean that the subjects simply indicated to the monitor the number of the test tube they were going to attempt to influence. At no time did the subjects have physical contact with the test tubes, because they were in the locked ice chest--to which the biological technician, alone, possessed the key.

The subjects were informed by the monitor that the set of nine test tubes, as with all biological systems, represented a natural spread of mutation rates, and that their task would most likely be facilitated if they tried to mentally influence the tubes in the direction they were already naturally inclined. A typical experimental session proceeded as follows:

- The subject chose whether he/she wished to begin the session with either the high-aim or low-aim condition.
- Having decided upon the condition, the subject was directed first to "select" (i.e., psychoenergetically identify) a test tube that already exhibited a mutation rate in the direction of the chosen condition, and then to mentally "promote" it (i.e., via some type of Remote Action mechanism) in that direction. For example, in the low-aim condition, a subject would be encouraged to "psychically scan" the session set of nine test tubes, and to select one that was predisposed toward producing a low mutation rate. The subject then attempted to mentally influence the biological culture in the tube, in an effort to further inhibit the mutation rate.
- For a given condition, a subject selected a tube and attempted to influence it for as long as he/she deemed necessary (usually 30 to 60 seconds), selected a second tube to concentrate on, then selected a third tube and concentrated on it. The strategies employed in the attempts at influencing the cultures were left to the subject's discretion. In the low-aim condition, for example, several subjects envisioned or "willed" that the *Salmonella* were dying, or would starve to death once they were plated in a histidine-free medium. In the high-aim condition, several subjects reported that they envisaged the bacteria dividing and multiplying at an extremely rapid rate, or that they were changing somehow into a new strain of bacteria that grew well in the absence of histidine.

- The subjects were encouraged to complete their efforts with all three test tubes in a given condition before moving on to the other condition, so as to avoid dividing their attention between the selection of high- and low-aim test tubes, and to maintain continuity of imagery strategies during the "influence" phases. The subjects were also admonished to avoid excessive strategy searching during the influence phase, in order to ensure that their attention would be focused on the actual task at hand rather than on the strategy employed.
- After having finished with three tubes in one condition, the subject was encouraged to take a brief rest. The same selection and concentration process was then carried out with three test tubes of the subject's choice in the other condition.

For every IDS unfavorable session, the protocol dictated *a priori* that Test Tubes 1, 2, and 3 (the first three tubes on the left as the subject faced the ice chest) were "low aim," Tubes 4, 5, and 6 were "no aim" or controls, and Tubes 7, 8, and 9 (the last three tubes on the right) were "high aim." The subject was specifically instructed to "mentally cause" a decreased mutation rate in Tubes 1, 2, and 3 and an increased mutation rate in Tubes 7, 8, and 9, and not to attempt to actively influence the mutation rates of Tubes 4, 5, and 6. A typical experimental session proceeded as follows:

- As in the case of the IDS favorable session, the subject chose whether he/she wished to begin the session with either the high-aim or low-aim condition.
- In the low-aim condition, the subject attempted to inhibit the mutation rates in Test Tubes 1, 2, and 3, one at a time and in the order of his choosing. Again, the subject determined the time of "concentration effort" per tube, which lasted from approximately 30 to 60 seconds. In the high-aim condition, the subject endeavored to increase the mutation rates in Tubes 7, 8, and 9, one at a time and in the order of his/her choosing. The strategies employed in these attempts at mentally influencing mutation rates were the same as those discussed for the IDS favorable session above.
- As in the IDS favorable condition, the subject was directed to complete the effort with all three test tubes in one condition before shifting attention to the other condition; a rest period between the two conditions was also encouraged.

The monitor's principal task during both IDS favorable and IDS unfavorable sessions was to fill out a *Bio-PK Form* (see Figure 1). Prior to start of the session, the "Session I.D.," "Viewer I.D.," and "Date" portions of the form were completed. Once the subject began his process of selection, a session "Start Time" was noted, and the test-tube selections were recorded by filling in the boxes (provided on the form's test-tube icons) with

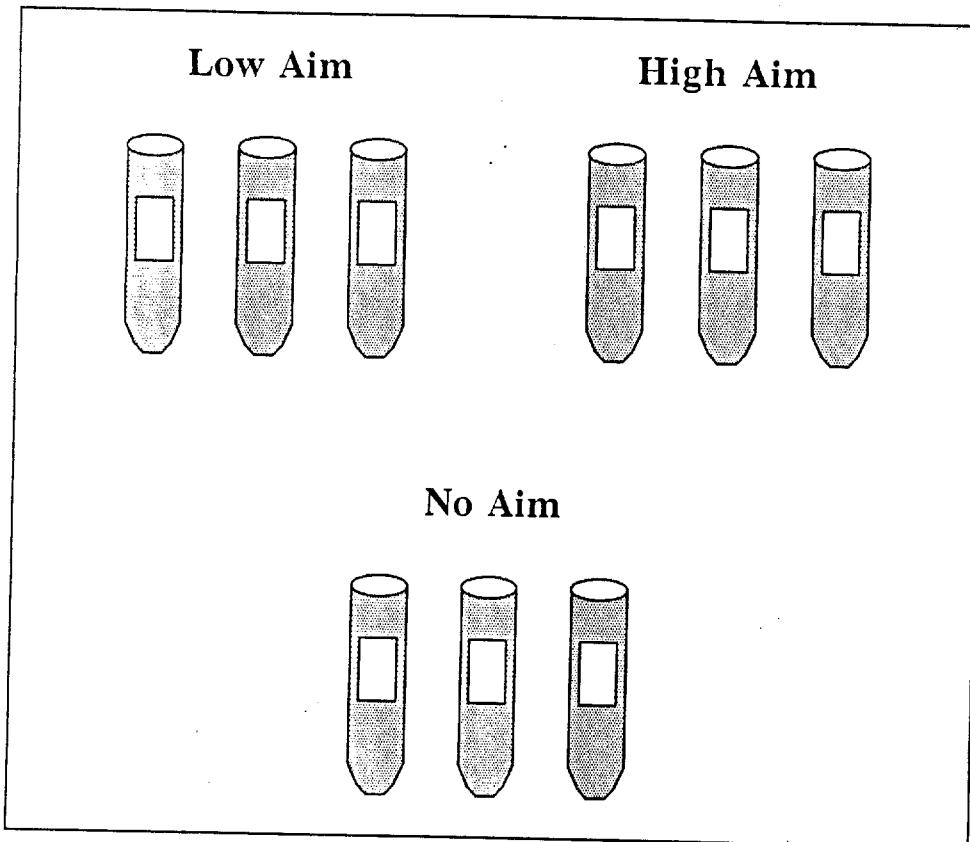
BIO - PK FORM

Session I.D. _____

Viewer I.D. _____

Date: _____

Start Time: _____



Comments:

FIGURE 1

SAMPLE BIO-PK FORM

the appropriate test-tube-selection numbers. The order of selection and an estimate of the duration of effort per each test tube were noted in the "Comments" section. Upon debriefing the subject at the end of a session, the monitor also recorded any comments the subject wished to make regarding possible strategies employed in the performance of the task, and any personal statements the subjects wished to volunteer pertaining to their state-of-mind, health, and so forth.

4. Postsession Protocols

At the conclusion of the session, and after the departure of the subject from the psychoenergetics facility, the monitor once again covered the ice chest, then transported it on the cart back to the Microbial Genetics Laboratory.

The microbiologist removed the nine bacterial cultures from the ice chest and placed them, together with the additional two *extrasession* control cultures, in an incubator (G24 Environmental Incubator Shaker, New Brunswick Scientific Company, Inc., Edison, New Jersey). The bacterial cultures were shielded from visible light by aluminum foil, and grown with gentle shaking (100 rpm) for about 24 hours.

Following the incubation period, testing of the eleven bacterial cultures to determine the extent of mutation induction was initiated. The testing was divided into two parts:

- Quantitation of number of cells plated, which measures the number of plated cells that are able to form colonies (CFU) on medium containing histidine (yeast complete medium).
- Quantitation of mutant cells, which measures the number of cells that are able to grow in the absence of histidine.

The quantitation of CFU was accomplished according to a standardized set of laboratory procedures. First, each of the eleven bacterial cultures (i.e., the cultures contained in the nine-session test tubes plus the two controls) was serially diluted by combining 0.20 ml of the culture with 1.80 ml of sterile saline until an overall million-fold dilution was obtained (10^{-6}). Complete medium plates were then divided into three sections with a marking pen, and a 10- μ l aliquot of the 10^{-4} , 10^{-5} , and 10^{-6} dilutions were then delivered in triplicate to the appropriate sections on the plate. The 10- μ l spots were allowed to dry on the surface of the solid medium in the plates. The plates were then incubated at 37°C for up to 24 hours

to allow for growth and appearance of colonies. The colonies were then counted manually using a New Brunswick Scientific Bactronic Colony Counter, Model C110, and the data were recorded for each dilution. In all cases, the 10^{-5} bacterial dilution provided an acceptable number of nonoverlapping individual colonies to back-calculate the total number of bacteria in 1 ml of the overnight culture.

The quantitation of the number of mutants (histidine independent cells) present in the overnight cultures was obtained according to the following set of procedures. First, a 100- μ l aliquot (approximately 10^8 cells) of the undiluted culture was combined with two ml of molten (43°C) top agar in a 13- \times -100-mm test tube. Vortexing of the cells in the top agar ensured even distribution of the cells when poured on the selective minimal glucose plates. Each culture was then plated in triplicate, and the plates were incubated at 37°C for 48 hours. When the incubation process was complete, revertant colonies were counted manually, and individual plate counts were recorded.

For each of the eleven bacterial cultures, an average CFU for 10 μ l of the 10^{-5} dilution was obtained and adjusted in terms of CFU per ml of the original culture. The mutation frequency was then calculated as

$$\text{mutation frequency}/10^6 \text{ cells} = \frac{\text{number mutant cells/ml}}{\text{CFU/ml}} \times 10^6$$

When all of the calculations for a given session were completed, the microbiologist provided the monitor with an experimental data sheet detailing the growth rates of the biological samples and the pertinent experimental parameters involved in the biological procedures (see Figure 2). The most salient column to note on this form is the "Mutants/ 10^6 Cells," which is the number of cells per million cells per test tube that were able to grow in the absence of histidine. The most desirable outcome of a session was obtained if the subject's "high-aim" test tubes exhibited the highest values from this column, and "low-aim" test tubes exhibited the lowest values.

After obtaining the form for a given session, the monitor completed a session feedback sheet (see Figure 3). The test tube numbers of the subject's choices for the IDS favorable condition (or the predetermined Numbers 1, 2, 3, 7, 8, and 9 for the IDS

Experiment: 104. sdInnoculation Date: 9/26/84 (wed) Time: 9:35 AM GM Broth Batch Date: 9/17/84Bacteria Stock Incubation Date: 9/16-9/27/84 Start Time: 10:30 AM End Time: 9:40 AM Total Time: 23.2 hrsSurvival (YC Plates) Incubation Start Time: 10:40 AM End Time: 10:09 AM (9/18) Total Time: 19.25 hrs Temp: 30°CMutation (Bio Plates) Incubation Start Time: 11:20 AM End Time: 9:30 AM (9/18) Total Time: 16.2 hrs Temp: 37°CBio Plate Batch Date: 9/17/84 YC Plate Batch Date: 9/28/84 Saline Batch Date: 9/25/84TA = 8/27/84

Tube #	Mutants/.100 ml	Aver. Mutant/.1 ml	>100	colonies/ 10 μ l			average cells/ml	mutants/ 10 ⁶ cells
				dil: 10 ⁻⁴	10 ⁻⁵	10 ⁻⁶		
1	53 69 62	61	>100	52 44 58	4 5	5	5.1×10^8	1.2
2	98 98 97	98	>100	25 37 26	4 2	4	2.9×10^8	3.4
3	84 92 91	89	>100	24 26 26	5 4	1	2.5×10^8	3.6
4	91 96 94	94	>100	19 22 22	0 3	4	2.1×10^8	4.5
5	99 96 90	95	>100	20 30 22	4 1	7	2.4×10^8	4.0
6	59 69 67	65	>100	33 43 38	2 4	7	3.8×10^8	1.7
7	76 76 96	83	>100	27 21 29	5 3	0	2.6×10^8	3.2
8	70 92 79	80	>100	23 23 24	2 6	3	2.3×10^8	3.5
9	95 124 106	108	>100	26 26 24	1 0	2	2.5×10^8	4.3
10	80 91 73	81	>100	35 30 40	1 4	5	3.5×10^8	2.3
11	91 104 83	93	>100	20 28 20	2 0	2	2.3×10^8	4.0

FIGURE 2

BACTERIAL ASSAY FORM SAMPLE

unfavorable condition) were written in the boxes below the appropriate low aim or high aim test-tube icons. The monitor then consulted the "Mutants/ 10^6 Cells" column on the form in order to rank, relative to each other, the mutation rates of the six chosen test-tube samples. To represent the results graphically, the boxes on the three-test tube icons corresponding to the three highest mutation rates were colored green, and the three boxes corresponding to the three lowest mutation rates were colored red. The monitor also colored the "High Mutation" box on the key green, and the "Low Mutation" box red, then filled in the appropriate session identification number. As mentioned above, the session feedback sheet was presented to the subject, whenever possible, just prior to the start of the subject's next session.

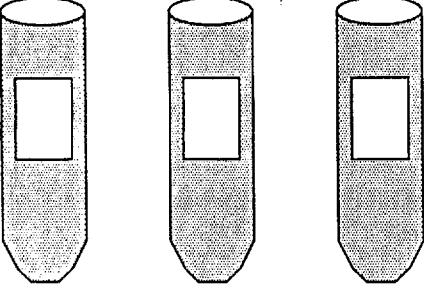
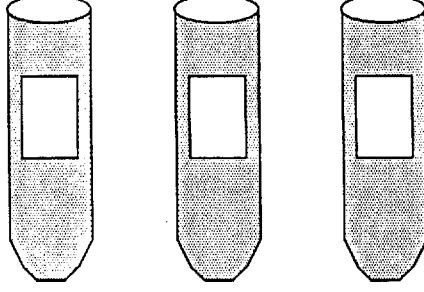
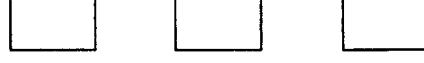
Session I.D. _____	Key
	<input type="checkbox"/> High Mutation
	<input type="checkbox"/> Low Mutation
Low Aim	High Aim
	
	

FIGURE 3

SAMPLE FEEDBACK FORM

IV RESULTS AND DISCUSSION

Seven subjects participated in six experimental sessions each: three for the IDS condition, and three for the RA condition. Of the total of 378 test tubes used in these sessions, 63 tubes corresponded to the high-, low-, and no-aim condition, respectively, for the two hypotheses under test. Thus, for a given hypothesis, there were 126 tubes to test for differential aim effects. An estimate of the number of trials necessary for a sensitive test was derived from published work, and was calculated on the basis of a significant t-test assuming a magnitude of effect on the order of 0.5σ . That is, it was assumed *a priori* that for either the RA or IDS hypothesis, individuals would be able to produce an effect of 0.5σ . Because of the large standard deviation (0.46 mutation cells per million) observed in 187 baseline control tubes (cf. Figure 4), it was hypothesized that three sessions per subject in both the RA and IDS conditions would be sufficient to demonstrate a significant effect.

Due to large "batch" variations (i.e., variation between sets of nine experimental test tubes), the normalization technique used by Nash was adopted. The measured mutation rate for each test tube was divided by the set-of-nine average and multiplied by the global average mutation rated for the 378 tubes in the experiment. Or,

$$\text{normalized mutation rate} = \text{mutation rate} \times \frac{\text{global average}}{\text{set average}}$$

Table 1 shows the normalized mutation rates observed during this experiment, summed across all subjects for the RA and the IDS conditions, respectively. Following the analysis done by Nash, pairs of t-tests between the various aims and conditions have been calculated. During the RA condition, there were no significant differences among the aims. When subjects were allowed to choose their test tubes, however, the low-aim condition was significantly lower than the no-aim condition ($p \leq 0.05$). While the low-high-aim difference was in the correct direction, the difference was not significant. There were no significant differences for various aims observed in the RA condition.

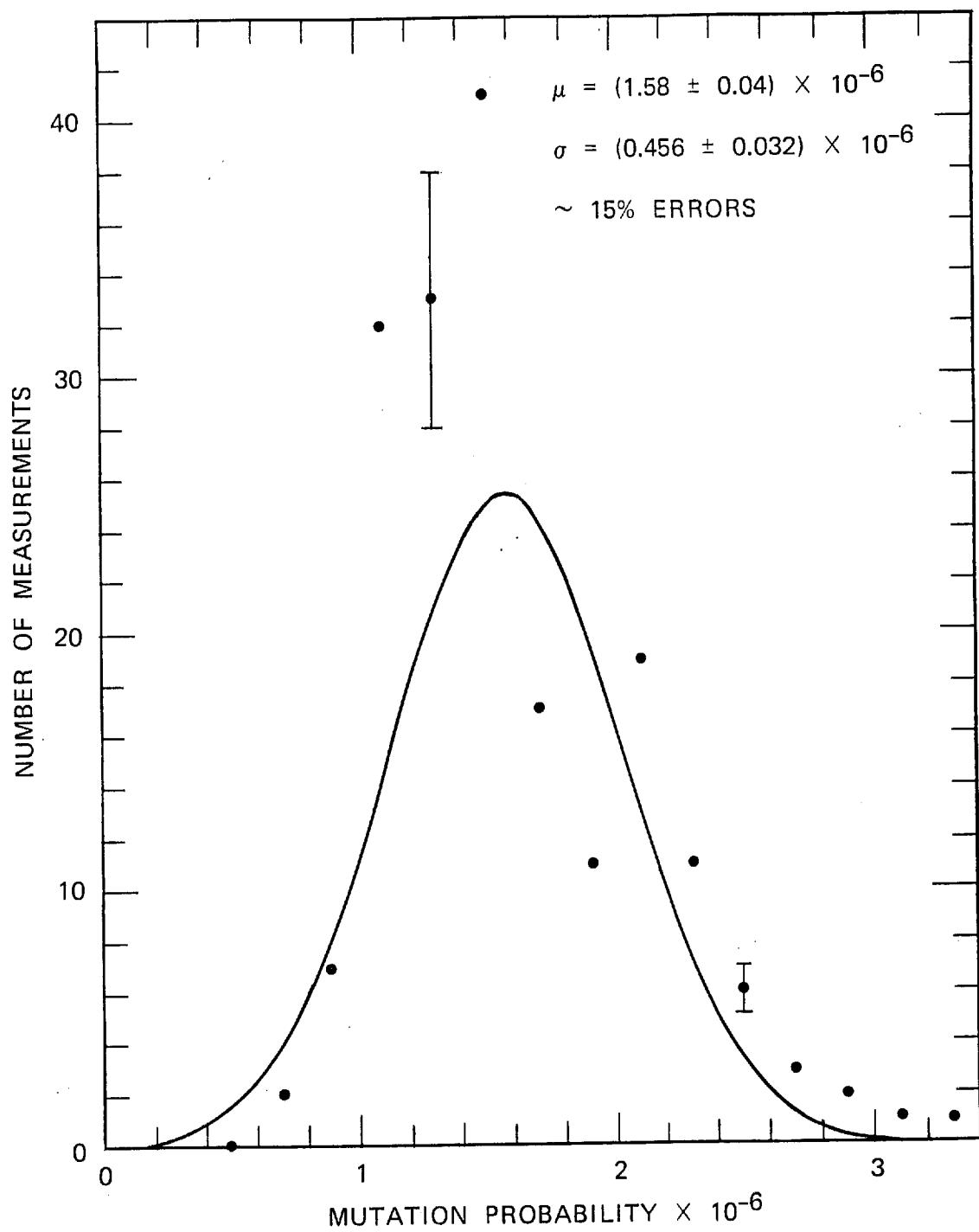


FIGURE 4 DISTRIBUTION OF MUTATION PROBABILITY FOR 187 BASELINE TEST TUBES

Table 1

NORMALIZED MUTATION RATES $\times 10^{-6}$ FOR ALL SUBJECTS

Condition	Aim		
	Low	No	High
Remote Action (RA)	3.08	3.17	3.06
Remote Action (RA)	3.37	2.92	2.88
Remote Action (RA)	3.08	2.97	3.13
Mean	3.18	3.02	3.02
Statistics*			
	t (Low < No) = -1.160	n.s.	
	t (No < High) = 0.412	n.s.	
	t (Low < High) = -1.200	n.s.	
Condition	Aim		
	Low	No	High
Intuitive Data Sorting (IDS)	2.80	3.25	3.23
Intuitive Data Sorting (IDS)	3.06	3.08	3.11
Intuitive Data Sorting (IDS)	3.11	3.12	2.90
Mean	2.99	3.15	3.08
Statistics*			
	t (Low < No) = 1.690	0.047†	
	t (No < High) = -1.190	n.s.	
	t (Low < High) = 0.550	n.s.	

* Degrees of freedom = 124.

† p-value.

From this analysis, it can be concluded that psychoenergetic functioning occurred that was consistent with results reported by Nash. While the size of the effect observed in this experiment was small compared with Nash's, it is consistent with the IDS hypothesis that certain subjects demonstrate a psychoenergetic ability to sort test tubes according to mutation rate. (It should be noted that an IDS channel was also present in Nash's experiment, but that Nash's significant result was interpreted exclusively within the RA hypothesis framework).

There was no compelling evidence, however, that subjects are able to interact causally with this particular biological system.

Tables 2 through 8 display the data contained in Table 1 in a subject-by-subject format. One subject (310) produced strong effects in the IDS condition: his low-aim condition was significantly lower than his high-aim condition ($p \leq 0.003$), and his low-aim condition was also significantly lower than the no-aim condition ($p \leq 0.018$).

(U) In addition to the various t-tests, a multiway analysis of variance (ANOVA) was conducted as a second form of analysis (post hoc). Aim (low, no, and high) and condition (IDS and RA) were used as the two "main effects" for the analysis. When the ANOVA examines one "main effect," it sums all the data in the other "main effects." For example, to examine the IDS and RA condition, the ANOVA sums across all aims. Likewise, to examine an aim effect, the ANOVA sums across the IDS and RA conditions. No significance was anticipated in these two dimensions, and none was observed. Significance was observed, however, in the interaction term between the IDS and the RA condition ($p \leq 0.05$), which may indicate that there is some difference between the IDS and RA conditions when examined as a function of aim. This does not imply that IDS or RA is "more significant." It should be noted that the interpretation of the ANOVA interaction term has been traditionally difficult, and Rosenthal has suggested that ANOVA with more than one "main effect" should not be used in the social/psychological sciences.* The analysis has been included here merely for the sake of completeness.

In summary, this experiment has produced a relatively weak, but statistically significant effect, which most readily supports the conclusion that subjects are able to acquire information, psychoenergetically, about the mutation rates of *Salmonella*, but are unable to cause physical perturbations in these bacteria. To reiterate the criteria set forth in the *Introduction*, a physical system will not be considered a candidate intrusion detector unless it registers energetic effects directly (as a result of intentional perturbation), or indirectly (as a result of concomitant acquisition of information). To first order, therefore, it must be concluded on the basis of this one experiment, that the *Salmonella* bacterium does not appear to be a promising intrusion detector.

* R. Rosenthal and R. Rosnow, *Essentials of Behavioral Research*, p. 254 (McGraw Hill Book Co., New York, 1984).

Table 2

NORMALIZED MUTATION RATES $\times 10^{-6}$
 (Subject 164)

Condition	Aim		
	Low	No	High
Remote Action (RA)	3.00	2.76	3.54
Remote Action (RA)	3.69	2.90	2.59
Remote Action (RA)	3.06	3.09	3.09
Mean	3.25	2.92	3.07
<hr/>			
Statistics*	t (Low < No) = -1.430 n.s.		
	t (No < High) = 0.484 n.s.		
	t (Low < High) = -0.475 n.s.		
<hr/>			
Condition	Aim		
	Low	No	High
Intuitive Data Sorting (IDS)	2.99	2.89	3.26
Intuitive Data Sorting (IDS)	2.79	3.54	2.93
Intuitive Data Sorting (IDS)	3.57	2.68	2.83
Mean	3.11	3.04	3.01
<hr/>			
Statistics*	t (Low < No) = -0.310 n.s.		
	t (No < High) = -0.112 n.s.		
	t (Low < High) = -0.475 n.s.		

* Degrees of freedom = 16.

Given the weak statistical nature of the effect and the potential operational importance of intrusion detection, replication is recommended for a variety of compelling reasons. First, there are a number of proposed methodological changes to this experiment (as discussed in Chapter V) that would in all likelihood enhance the robustness of the effect. From this perspective, the experiment might legitimately be considered a pilot study. Second, this is the first experiment of its kind that has used *Salmonella* as the target biological system;

Table 3

NORMALIZED MUTATION RATES $\times 10^{-6}$
(Subject 240)

Condition	Aim		
	Low	No	High
Remote Action (RA)	2.91	3.45	2.87
Remote Action (RA)	3.48	3.09	2.68
Remote Action (RA)	3.45	2.58	3.16
Mean	3.28	3.04	2.90
 Statistics*			
t (Low < No) = -0.809 n.s.			
t (No < High) = -0.543 n.s.			
t (Low < High) = -1.443 n.s.			
Condition	Aim		
	Low	No	High
Intuitive Data Sorting (IDS)	2.59	3.23	3.40
Intuitive Data Sorting (IDS)	3.40	2.87	2.98
Intuitive Data Sorting (IDS)	2.98	3.20	2.92
Mean	2.99	3.10	3.10
 Statistics*			
t (Low < No) = 0.345 n.s.			
t (No < High) = -0.004 n.s.			
t (Low < High) = 0.317 n.s.			

* Degrees of freedom = 16

thus, at least one attempt at replication would be appropriate, before any definitive statements could be made regarding the efficacy of *Salmonella* as an intrusion detector. Finally, the statistically significant result in the IDS condition--the informational ability to obtain a desired outcome using intuition--demonstrates the existence of a psychoenergetic ability that has numerous operational applications; replications could be designed to further explore and enhance this potentially useful psi talent.

Table 4

NORMALIZED MUTATION RATES $\times 10^{-6}$
(Subject 310)

Condition	Aim		
	Low	No	High
Remote Action (RA)	2.73	2.84	3.74
Remote Action (RA)	3.21	2.91	2.99
Remote Action (RA)	2.53	3.16	2.99
Mean	2.82	2.97	3.32
Statistics*	t (Low < No) = 0.394	n.s.	
	t (No < High) = 0.976	n.s.	
	t (Low < High) = 1.485	n.s.	
Condition	Aim		
	Low	No	High
Intuitive Data Sorting (IDS)	2.71	3.36	3.04
Intuitive Data Sorting (IDS)	2.65	2.99	3.47
Intuitive Data Sorting (IDS)	2.64	3.06	3.38
Mean	2.67	3.14	3.29
Statistics*	t (Low < No) = 2.298	0.018 †	
	t (No < High) = 0.677	n.s.	
	t (Low < High) = 3.294	0.003 †	

* Degrees of freedom = 16.

† p-value.

Table 5

NORMALIZED MUTATION RATES $\times 10^{-6}$
(Subject 385)

Condition	Aim		
	Low	No	High
Remote Action (RA)	3.21	3.28	2.81
Remote Action (RA)	3.57	2.62	2.89
Remote Action (RA)	3.08	2.92	3.28
Mean	3.27	2.95	2.99
Statistics*			
t (Low < No) = -0.948 n.s.			
t (No < High) = 0.144 n.s.			
t (Low < High) = -1.012 n.s.			
Condition	Aim		
	Low	No	High
Intuitive Data Sorting (IDS)	2.75	3.02	3.26
Intuitive Data Sorting (IDS)	2.86	2.97	3.30
Intuitive Data Sorting (IDS)	3.21	3.29	2.68
Mean	2.94	3.09	3.08
Statistics*			
t (Low < No) = -0.310 n.s.			
t (No < High) = -0.052 n.s.			
t (Low < High) = 0.487 n.s.			

* Degrees of freedom = 16.

Table 6

NORMALIZED MUTATION RATES $\times 10^{-6}$

(Subject 531)

Condition	Aim		
	Low	No	High
Remote Action (RA)	2.75	3.18	3.37
Remote Action (RA)	3.17	2.95	2.95
Remote Action (RA)	3.54	2.70	2.91
Mean	3.15	2.86	3.08
 Statistics*			
t (Low < No) = -1.494 n.s.			
t (No < High) = 1.059 n.s.			
t (Low < High) = -0.333 n.s.			
Condition	Aim		
	Low	No	High
Intuitive Data Sorting (IDS)	2.94	3.96	3.30
Intuitive Data Sorting (IDS)	3.19	2.84	3.19
Intuitive Data Sorting (IDS)	2.96	3.52	2.82
Mean	3.03	3.44	2.77
 Statistics*			
t (Low < No) = 1.281 n.s.			
t (No < High) = -2.119 0.050†			
t (Low < High) = -1.123 n.s.			

* Degrees of freedom = 16.

† p-value; 2-tailed

Table 7

NORMALIZED MUTATION RATES $\times 10^{-6}$
(Subject 807)

Condition	Aim		
	Low	No	High
Remote Action (RA)	3.57	3.57	2.21
Remote Action (RA)	3.02	2.90	3.30
Remote Action (RA)	2.86	2.80	3.53
Mean	3.15	3.09	3.01
Statistics*	$t (\text{Low} < \text{No}) = -0.202$ n.s. $t (\text{No} < \text{High}) = -0.242$ n.s. $t (\text{Low} < \text{High}) = -0.440$ n.s.		
Condition	Aim		
	Low	No	High
Intuitive Data Sorting (IDS)	3.11	3.09	2.94
Intuitive Data Sorting (IDS)	2.86	3.30	3.06
Intuitive Data Sorting (IDS)	3.46	3.00	2.49
Mean	3.14	3.13	2.83
Statistics*	$t (\text{Low} < \text{No}) = -0.053$ n.s. $t (\text{No} < \text{High}) = -1.332$ n.s. $t (\text{Low} < \text{High}) = -1.322$ n.s.		

* Degrees of freedom = 16.

Table 8

NORMALIZED MUTATION RATES $\times 10^{-6}$
(Subject 997)

Condition	Aim		
	Low	No	High
Remote Action (RA)	3.37	3.09	2.88
Remote Action (RA)	3.46	3.06	2.77
Remote Action (RA)	2.76	3.56	2.96
Mean	3.20	3.12	2.87
 Statistics*			
t (Low < No) = -0.248 n.s.			
t (No < High) = -0.857 n.s.			
t (Low < High) = -1.319 n.s.			
Condition	Aim		
	Low	No	High
Intuitive Data Sorting (IDS)	2.51	3.18	3.41
Intuitive Data Sorting (IDS)	3.46	2.82	2.87
Intuitive Data Sorting (IDS)	2.94	3.06	3.15
Mean	2.97	3.02	3.15
 Statistics*			
t (Low < No) = 0.167 n.s.			
t (No < High) = 0.640 n.s.			
t (Low < High) = 0.591 n.s.			

* Degrees of freedom = 16.

V SUGGESTIONS FOR FUTURE STUDIES

In this section we provide a discussion of the difficulties that were encountered in the course of completing this experiment, and the suggested protocol modifications for follow-on experiments.

A. The Role of Feedback

The role of feedback to the subject regarding his/her performance is generally believed to be essential for optimizing the performance of psychoenergetic tasks. In the vast majority of experiments performed at SRI, feedback is administered promptly, and is typically visual in nature. In remote viewing experiments, for example, feedback usually consists of a pictorial representation of the target site in question, which is presented to the subject immediately at the conclusion of the trial. In random number generator (RNG) experiments, feedback has typically taken the form of an "on-line" random walk video display, representing the output of either a radioactive source or a noise diode.

Because of the time factors inherently involved in growing and plating the bacterial cultures, feedback to the subject in this experiment was minimally delayed by a period of 48 hours. In many cases, the delay was considerably longer than this because of scheduling difficulties involving the subjects or the technician. It may not be possible to circumvent this feedback-delay problem when using bacteriological systems, but in the absence of definitive knowledge concerning the necessity for feedback, it should be recognized that delayed feedback may inhibit psychoenergetic performance. At the very least, feedback should probably be provided to the subject at fixed, rather than at variable intervals, so that the expectation of feedback can be fulfilled on a known and constant schedule.

The type of feedback that is administered should also be subject to careful consideration. As mentioned above, Figure 3 (Section III.3) is an example of the feedback sheet that was used for this experiment. To indicate the subject's result graphically, the boxes on the three test-tube icons corresponding to the three highest mutation rates were colored green, and the three boxes corresponding to the three lowest mutation rates were colored red.

This form may prove to be too abstract to provide good feedback; other modes might be considered, that would better concretize the experience for the subject. For example, in future experiments, the subject might be shown iconic representations of agar plates: many spots on these "plates" would correspond to many bacterial colonies and hence a high mutation rate; few spots would represent the inverse situation. Or, perhaps the subject could be shown the actual plates with their bacterial cultures--if there is enough pertinent feedback information to be gleaned from such an experience.

B. **Screening Criteria and Presentation of Psychoenergetic Task**

For the experiment detailed in this document, subjects were chosen primarily on the basis of general availability, and on an expressed interest in psychoenergetic research. Because the SRI program has recently implemented the Psychological Assessment System (PAS) in conjunction with other psychoenergetic projects (e.g., remote viewing enhancement), it is suggested that this system be used to select subjects for follow-on biological experiments. Thus, the individuals who participated in the study described here could be psychologically profiled according to the PAS, and the profiles of those who performed successfully could be used as prescriptive templates for selecting new participants for future experiments. Subject 310, for example, demonstrated significant ability in sorting test tubes according to the IDS hypothesis; it is suggested that his PAS profile be used, in part, to select subjects for future psychoenergetic experiments involving biological systems.

Another variable that should be considered in subject selection involves the degree to which they possess an *a priori* understanding of the actual biological "mechanics" of bacteriological systems. None of the subjects selected for the study presented here were biologists *per se*, nor were they particularly familiar with microbial genetics: it seemed, therefore, somewhat difficult at times to impart to the subject an understanding of the causes of biological mutations and how such mutations might be created psychoenergetically. In future studies it might be instructive, to explore the degree to which comprehensive knowledge concerning the biological aspects of the experiment promotes or inhibits successful performance of the psychoenergetic task.

C. Biological Protocols

For any given trial, the technician should randomize the nine inoculated test tubes prior to placing them in the rack. This will ensure that ordering effects caused by sequential pipetting will not be evidenced in the IDSU or RA condition.

D. Future Experiments with Other Biological Systems

It is difficult to interpret the absence of a detectable RA effect in the experiment reported in this document. One possible interpretation is that the *Salmonella typhimurium* are simply one kind of biological system that is not susceptible to psychoenergetic influence. Given that this hypothesis is verified in further experiments with *Salmonella*, a future approach would be to examine RA in relation to other biological systems.

1. Single-Cell Systems

Examples of other single-cell biological systems that could be used to study RA are listed below:

- **Bacterial Chemotaxis**--The extent of reversion of nonmotile mutant bacteria to ability to move towards a gradient of a chemical attractant would be measured. Microscopic examination of the cells could be used to monitor the reversion. Time-lapsed photography could also be used to obtain a permanent record of the observations made during the experiment.
- **Selection of UV (ultraviolet light) Sensitive Bacterial Mutants**--RA-treated cell cultures would be allowed to grow to stationary phase, at which time their UV sensitivity would be determined by performing UV survival experiments.
- **Mammalian Cell Transformation**--Cultured cancerous cells are not contact inhibited; they grow in an irregular, randomly-oriented fashion in culture dishes. This is in contrast to cultured healthy normal cells, which are contact inhibited, and which form a monolayer in a neatly oriented fashion. When mouse C3H 10T-1/2 cells are cultured, some cells will spontaneously transform into cancerous cells. The transformation event can be increased by exposing the cells to physical or chemical agents. The extent of transformation is measured by determining how many transformed cells gave rise to visible foci that consist of randomly-oriented cells growing in multilayers, which appear as a very dense area (1-to-2-mm diameter).
- **Yeast Aneuploidy**--Malsegregation of chromosomes occurs spontaneously, or is induced by exposure of cells to physical or chemical agents. There are

a number of aneuploidy systems that would lend themselves to the effect of RA on aneuploidy. The advantage of using yeast organisms is their ease of handling, and the relatively short turnaround time (about one week) in obtaining results.

- **Molecular Biology and Mutation Induction**--At the molecular level, a study could be made of the effect of RA on mutation induction by either analyzing DNA sequences in bacterial/mammalian cells or by analyzing the amino acid sequence in proteins. This approach might prove to be a more efficient way of obtaining results in terms of time and labor, compared with the more conventional mutagenesis assays that rely on the phenotypic expression of the mutation.

In the not too distant future, it might well be possible to also study the effect of RA on oncogene activation in cultured mammalian cells.

2. Multicell Systems

In practical terms, the common fruit fly known as *Drosophila melanogaster* is most likely the only multicell system that could be used to study RA. The end point of such experiments would probably be changes in the eye color, which would reflect specific chromosomal mutations.

These are just a few representative examples of other types of biological systems that could be used in future RA/IDS experiments. Because many insights into protocol improvements have been obtained from our pilot experiment, it is our recommendation that the suggested methodological changes be incorporated into a formal replication study using *Salmonella*, again, as the target system, before experiments are undertaken with other biological systems for which the psychoenergetic protocols are not as well understood.

Appendix

BIOLOGICAL MATERIALS

The following biological materials were used for this experiment:

- Dispensers, 5 ml, sterile
- 13- \times -100-mm test tubes, sterile
- 16- \times -150-mm test tubes, with caps, sterile
- Physiologic saline, prepared according to SOP 374.412.3.6
- Glucose minimal (GM) broth, prepared according to SOP 374.412.3.14
- Top agar, prepared according to SOP 374.412.3.8.1
- Selective minimal medium plates, prepared according to SOP 374.412.3.12
- Complete medium (yeast complete) plates, prepared according to SOP 374.412.3.5
- Pipettes, sterile.